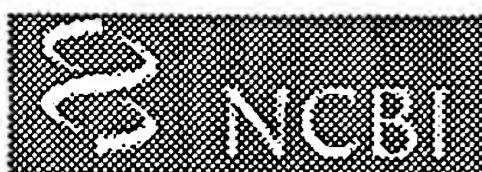


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S6	284779	recombination or recombinase\$ or restriction or ligat\$ or ligase\$	US-PGPUB; USPAT; EPO; DERWENT	OR	OFF	2005/08/02 11:25
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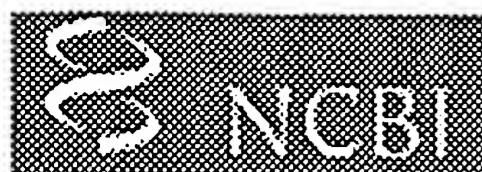
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Rapid construction of recombinant DNA by the univector plasmid-fusion system.

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Department of Biochemistry and Molecular Biology, Howard Hughes Medical Institute, Baylor College of Medicine, Houston, Texas 77030, USA.

PMID: 11075365 [PubMed - indexed for MEDLINE]

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The univector plasmid-fusion system, a method for rapid construction of recombinant DNA without restriction enzymes.

Liu Q, Li MZ, Leibham D, Cortez D, Elledge SJ.

Howard Hughes Medical Institute Verna and Marrs McLean Department of Biochemistry
Baylor College of Medicine One Baylor Plaza Houston Texas 77030 USA.

Background: Modern biological research is highly dependent upon recombinant DNA technology. Conventional cloning methods are time-consuming and lack uniformity. Thus, biological research is in great need of new techniques to rapidly, systematically and uniformly manipulate the large sets of genes currently available from genome projects. **Results:** We describe a series of new cloning methods that facilitate the rapid and systematic construction of recombinant DNA molecules. The central cloning method is named the univector plasmid-fusion system (UPS). The UPS uses Cre-lox site-specific recombination to catalyze plasmid fusion between the univector - a plasmid containing the gene of interest - and host vectors containing regulatory information. Fusion events are genetically selected and place the gene under the control of new regulatory elements. A second UPS-related method allows for the precise transfer of coding sequences only from the univector into a host vector. The UPS eliminates the need for restriction enzymes, DNA ligases and many in vitro manipulations required for subcloning, and allows for the rapid construction of multiple constructs for expression in multiple organisms. We demonstrate that UPS can also be used to transfer whole libraries into new vectors. Additional adaptations are described, including directional PCR cloning and the generation of 3' end gene fusions using homologous recombination in *Escherichia coli*. **Conclusions:** Together, these recombination-based cloning methods constitute a new comprehensive approach for the rapid and efficient generation of recombinant DNA that can be used for parallel processing of large gene sets, a feature that will facilitate future genomic analysis.

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